



## National Capital Region Network- Inventory and Monitoring Program

### *A New Record of Ephydatia muelleri in Prince William Forest Park (PRWI)*

Natural Resource Technical Report NPS/NCRN/NRTR—2010/363



**ON THE COVER**

Freshwater Sponge : *Ephydatia muelleri* in South Fork Quantico Creek at PRWI  
Photograph by: Tonya Watts NCRN I&M Program

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## *A New Record of Ephydatia muelleri in Prince William Forest Park (PRWI)*

Natural Resource Technical Report NPS/NCRN/NRTR—2010/363

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## Abbreviations and Acronyms

μS/cm	microSiemens per centimeter
ANC	acid neutralizing capacity
DC	District of Columbia
EMu	Electronic Museum System
ft	feet
I&M	Inventory and Monitoring Program
MD	Maryland
Mg/L	milligrams per liter
mL	milliliter
N	normal
NCRN	National Capital Region Network
NMNH	National Museum of Natural History
PRWI	Prince William Forest Park
SEM	scanning electron microscope
VA	Virginia
YSI	Yellow Springs Instruments

## Executive Summary

In 2007, during routine monitoring of South Fork Quantico Creek in Prince William Forest Park (PRWI), a freshwater sponge (*Porifera*) was observed. Several sponges were observed again in 2009 during routine monitoring along the same section of South Fork Quantico Creek and at its confluence with North Fork Quantico Creek.

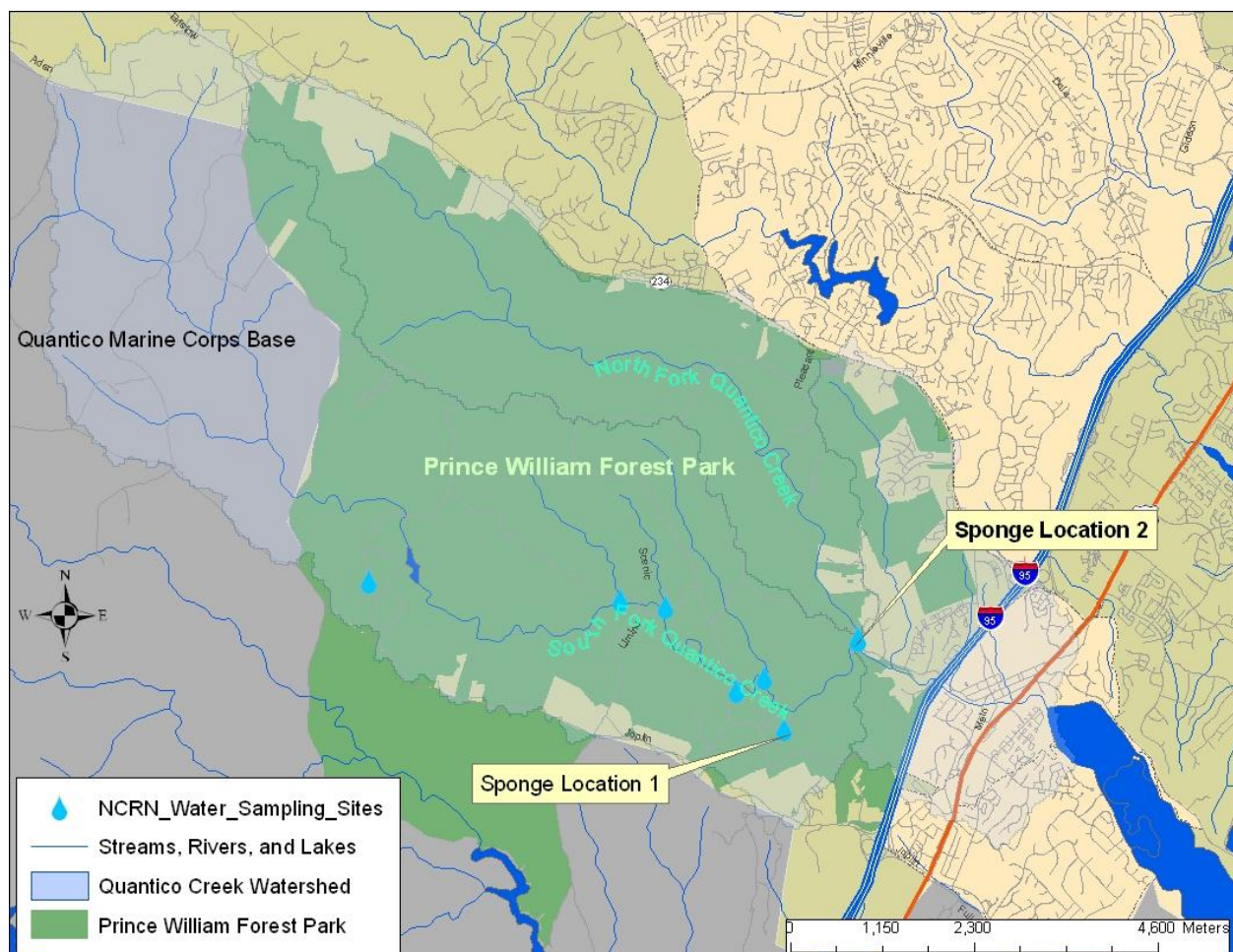
A search of National Park Service records did not find a recent occurrence of freshwater sponges within the boundaries of National Capital Region parks. A search of literature yielded an unidentified species in Pimmit Run between George Washington Memorial Parkway and the Potomac River in October 1974 and another unidentified species in Wolf Trap Creek at Wolf Trap National Park for the Performing Arts in 1975. In Virginia, Maryland or Washington, DC there have been few observations and even fewer documented specimens.

Sponges cannot be identified macroscopically. Identification is based on microscopic characteristics. The sponge samples from South Fork Quantico Creek were identified as *Ephydatia muelleri*, as first described by Lieberkuhn in 1855, by Klaus Ruetzler, Curator of *Porifera* at the Smithsonian Institution National Museum of Natural History (NMNH) Department of Invertebrate Zoology.

As part of the National Capital Region Network (NCRN) Inventory and Monitoring (I&M) Program, monthly water quality and quantity data are collected for each 1<sup>st</sup> to 3<sup>rd</sup> order perennial stream in PRWI. Data from the NCRN Program and other data sources for the park were examined with respect to expected ecological ranges for freshwater sponges.

## Introduction

In 2007 during routine monitoring of South Fork Quantico Creek in Prince William Forest Park (PRWI), a freshwater sponge (*Porifera*) was observed. Sponges were observed again in 2009 during routine monitoring along the same section of South Fork Quantico Creek (Sponge Location 1 in Figure 1) plus an additional location at its confluence with North Fork Quantico Creek (Sponge Location 2).



**Figure 1: Water monitoring sites and sponge locations in Prince William Forest Park and Quantico Creek Watershed**

A search of literature found few occurrences of freshwater sponges within the boundaries of National Capital Region parks. These occurrences are an unidentified species in Pimmit Run between George Washington Memorial Parkway and the Potomac River in October 1974 and another unidentified species in Wolf Trap Creek at Wolf Trap National Park for the Performing Arts in 1975 (1979 data from George Mason University, see Table 1). In Virginia, Maryland and Washington, D.C. there are few documented specimens (Smithsonian Institution holdings, see table 2).

**Table 1: Freshwater Sponges in Fairfax County, Virginia** (Cheryl Bright, pers. comm. 28 October 2009)

STREAM	LOCATION	DATE	NOTES
Pimmit Run	between the George Washington Memorial Parkway & Potomac River	11 Oct 1974	
Sugarland Run	at Route 606	2 Jul 1976	
Mine Run Branch	at River Bend Road	18 Jun 1976 28 Aug 1975	
Colvin Run	at Hunter Mill Road	5 Jun 1975	very common
Captain Hickory Run	at Walker Road	5 Jun 1975	common
Wolf Trap Creek	at Wolf Trap Park	10 Jun 1975	
Potomac River	at the mouth of Little Hunting Creek	27 Aug 1975	

**Table 2: MD, DC, and VA specimens of the family Spongillidae dating from the 1800s, found in the Smithsonian Institution EMu (Electronic Museum System)** (Cheryl Bright, pers. comm. 29 October 2009)

ACCESSION NUMBER	GENUS SPP.	CITY / STATE	WATERBODY	LATITUDE	LONGITUDE
371809	<i>Ephydatia japonica</i>	MD	Potomac River	38.416	79.6
338947	<i>Ephydatia muelleri</i>	Washington, DC	Potomac River	37.93889	-76.25278
35186	<i>Heteromeyenia ryderi</i>	VA	Dismal Swamp		
371809	<i>Spongilla aspinosa</i>	Norfolk, VA	Deep Creek Basin		
371809	<i>Heteromeyenia ryderi</i>	VA	Dismal Swamp		
178301	<i>Spongilla lacustris</i>	MD	Pomonkey Creek		
338947	<i>Ephydatia crateriformis</i>	Washington, DC	Potomac River	38.895	-77.03667
54100	<i>Ephydatia muelleri</i>	VA	Dyke (not Dyke Marsh)	37.5	-80.1
178301	<i>Ephydatia crateriformis</i>	Washington, DC	Potomac River	38.416	79.6

## **Setting**

PRWI is located in Prince William and Stafford Counties, Virginia. It straddles the Piedmont and Coastal Plain Physiographic Provinces resulting in the typical undulating landscape (rolling hills) of the Piedmont topped by the sandy soils of the Coastal Plain. The park also occurs within the transition zone between northern and southern climates, which supports many species to the limits of their ranges. The intersection of these characteristics creates a unique diversity of habitat, vegetative communities, and species composition not generally found in any single forest ecosystem. As the third largest unit of the National Park System in Virginia, the park also represents one of the largest protected parcels of undeveloped land within the Washington DC metropolitan area and serves as the largest piedmont forest ecosystem within the National Park System (NPS 2006).

PRWI is located within the Lower Potomac River drainage basin (USGS hydrologic unit 02070011). Two stream systems run through the park, Quantico Creek and Chopawamsic Creek, and eventually empty into the Potomac River. Numerous intermittent and perennial tributaries exist wholly on parkland and empty into these two systems. Historically, land use within these two systems has been agricultural or industrial in nature. Today, both watersheds are primarily forested, but contain some military-related land use.

The 30 square mile Quantico Creek watershed (Figure 1) is comprised of two streams, North Fork Quantico Creek and South Fork Quantico Creek, and numerous tributaries. The headwaters of North Fork Quantico Creek occur within PRWI and the stream runs through two former mining sites and two small man-made impoundments. Nearly 90% of the 7 square miles of the North Fork Quantico Creek watershed is protected by the park. The headwaters of South Fork Quantico Creek lie within Marine Corps Base Quantico, encompassing approximately 9 square miles. The stream enters the park where it runs through a moderately sized man-made impoundment. Two additional small man-made impoundments can be found on tributaries to South Fork. Roughly 10 square miles of South Fork Quantico Creek's watershed is contained within PRWI. The remaining 4 square miles of the Quantico Creek Watershed are in private ownership and primarily occur beyond the confluence of North Fork and South Fork along Quantico Creek (proper) (Petersen 2005). Protection of the Quantico Creek watershed is included in the park's enabling legislation.

Freshwater sponges are found in water bodies with high quality water and low levels of pollutants, disturbance and silt (Holley 2009). The presence of the organisms may indicate that South Fork Quantico Creek is a high quality stream. Determining the implications for Quantico Creek water quality management required taxonomic identification of the organism.

## **Methods**

### **Collection**

In the fall of 2009 a collection permit from PRWI was obtained ( #PRWI-2009-SCI-0019). Sponge samples were then collected from two points at South Fork Quantico Creek. Samples were carefully scraped from the rocks and put into 125ml sample bottles containing approximately 20 ml of 70% alcohol.

Removing a portion of the sponge is not harmful because they easily regenerate. Sponges cannot be identified macroscopically. Identification is based on microscopic spicule appearance; particularly gemmoscleres, a type of spicule that coats the gemmule (Thorp and Covich 2010). Gemmules are small, hard, round balls used for reproduction. They are easily seen with the naked eye. In late November 2009, when the samples were collected, most of the sponges had formed gemmules.

### **Identification**

Klaus Ruetzler, Curator of *Porifera* at the Smithsonian Institution NMNH Department of Invertebrate Zoology, prepared slides for the compound microscope and photographed the spicules through a Scanning Electron Microscope (SEM) (see Figure 2). The spicules are siliceous and are cleaned by boiling tissue in fuming nitric acid, then washed in changes of water and alcohol (in a test tube) and dried onto a glass slide, then covered by Permount and a cover slip (Ruetzler, pers. comm., 28 October 2009).

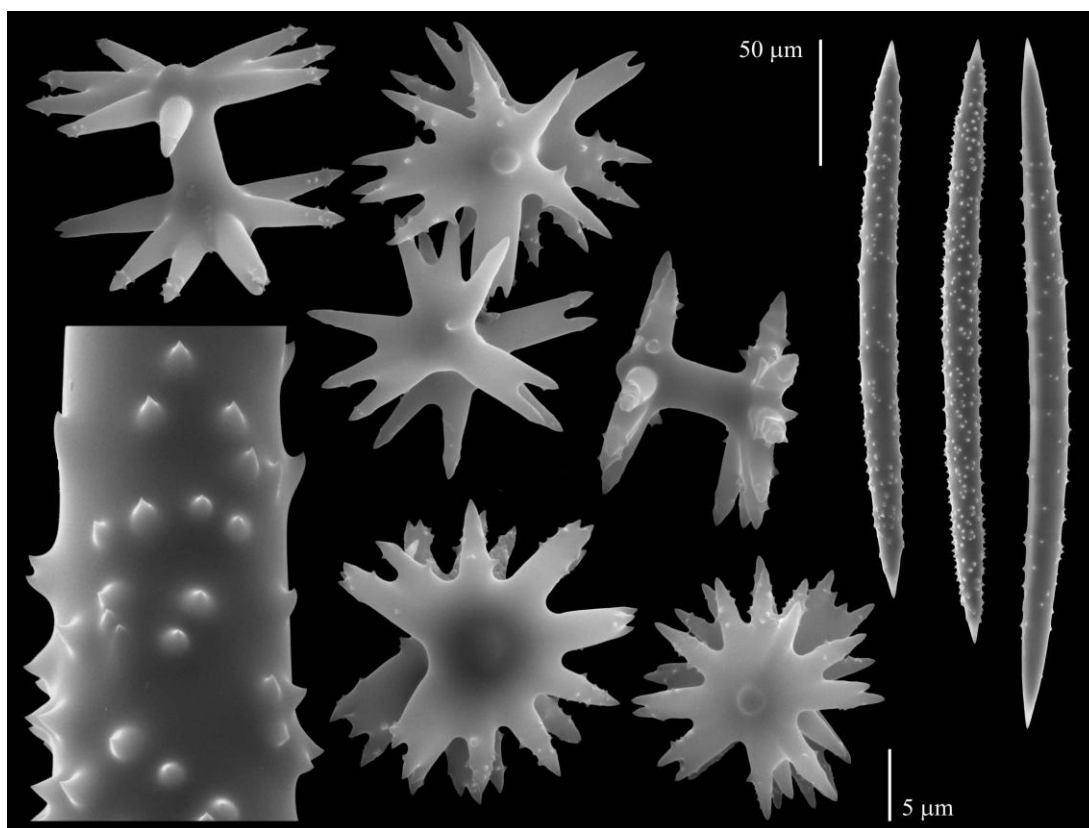
### **Water Quality and Quantity**

As part of the NCRN I&M Program monthly water quality and quantity data are collected for each 1<sup>st</sup> to 3<sup>rd</sup> order perennial stream in PRWI. Nutrient sample bottles are soaked in 6 N hydrochloric acid for 24-48 hours and rinsed in deionized water. A 125 ml sample is collected for analysis of nitrate and total phosphorous. A 500 ml sample is collected for analysis of acid neutralizing capacity (ANC). The ANC sample bottles are not washed with acid solution. During sample collection each bottle is rinsed three times with stream water and the fourth sample is stored in a cooler. All samples are kept refrigerated and analyzed in the water laboratory at the National Park Service, Center for Urban Ecology within 24-48 hours. Temperature, pH, salinity, dissolved oxygen (% and mg/L), specific conductance and conductivity are measured *in situ* with a Yellow Springs Instruments (YSI) Professional Plus. Wetted width, average depth, average flow, and discharge are recorded using a Sontek Flowtracker (Norris et al. 2008). Data from the NCRN Program and other data sources for the park were examined with respect to expected ecological ranges for freshwater sponges.

## Results

### Identification

The sponge samples from South Fork Quantico Creek were identified as *Ephydatia muelleri* - Lieberkuhn 1855, based on the microscopic gemmule and spicule shape (Figure 2) and agreement with the monograph by Penny & Racek (1968: pp. 85-87, Plate 7). *E. muelleri* is commonly referred to as “Mueller’s freshwater sponge.”



**Figure 2: Electron Micrograph of spicules of *Ephydatia muelleri* found in PRWI. Middle: gemmoscleres, left and right: megascleres.** Photos of PRWI sample courtesy of Klaus Ruetzler, Smithsonian Institution.

### Field Observations

South Fork Quantico Creek in PRWI is a 3<sup>rd</sup> order stream. Its depths vary from ankle deep to un-wadeable (approx 0.5-5.0 ft.). *Ephydatia muelleri* were originally found in the park in South Fork Quantico Creek. The first observation was just downstream of parking lot A and the other was about 2.7 km (1.67 miles) further downstream near the confluence of North Fork Quantico Creek. The two areas where they were found were light riffle and shallow pool areas. Both sponge populations were located at the transition zone of the piedmont and coastal plain regions. The sponges are encrusted on submerged rocks in the stream (Figure 3). They are bright green, opaque white or dull yellow in color. Visually, they can easily be mistaken for algae or green leaves in the stream. *In situ* the best way to determine if the organism is a sponge is by touch. They are coarse, but not rigid, pliable and have a hairy texture. They are not usually slimy like algae. They were first observed in South Fork Quantico Creek downstream of parking lot A during the summer of 2007 by NCRN water monitoring personnel. Sponges were not observed



again until the spring of 2009. Observations in summer and fall of 2009 suggest that the sponges may be growing and spreading downstream in South Fork Quantico Creek to the confluence with North Fork Quantico Creek. According to Smith (2001) wind, insects, birds, and mammals probably transport the resistant gemmules overland. Because they can grow on other organisms, they are also easily transported through the water (Smith 2001).



Figure 3: *Ephydatia muelleri* in South Fork Quantico Creek



In October 2009 the sponges were still green (Figure 4, left image), indicating the retention of a symbiotic algae. In November, some of them began to lose their skeleton and vibrant green color faded to pale yellow-brown. Most of the sponges had formed visible gemmules in a clear, resistant coat of collagen (Figure 4, right image). The water was too high and turbid in December 2009 and January 2010 to observe any remaining dormant sponges. New sponges were developing in April 2010.



Figure 4: Vibrant green *E.muelleri* encrusted on rocks, October 2009 (on right) and loss of skeletal mass and formation of gemmules, November 2009 (on left).

## Water Quality and Quantity

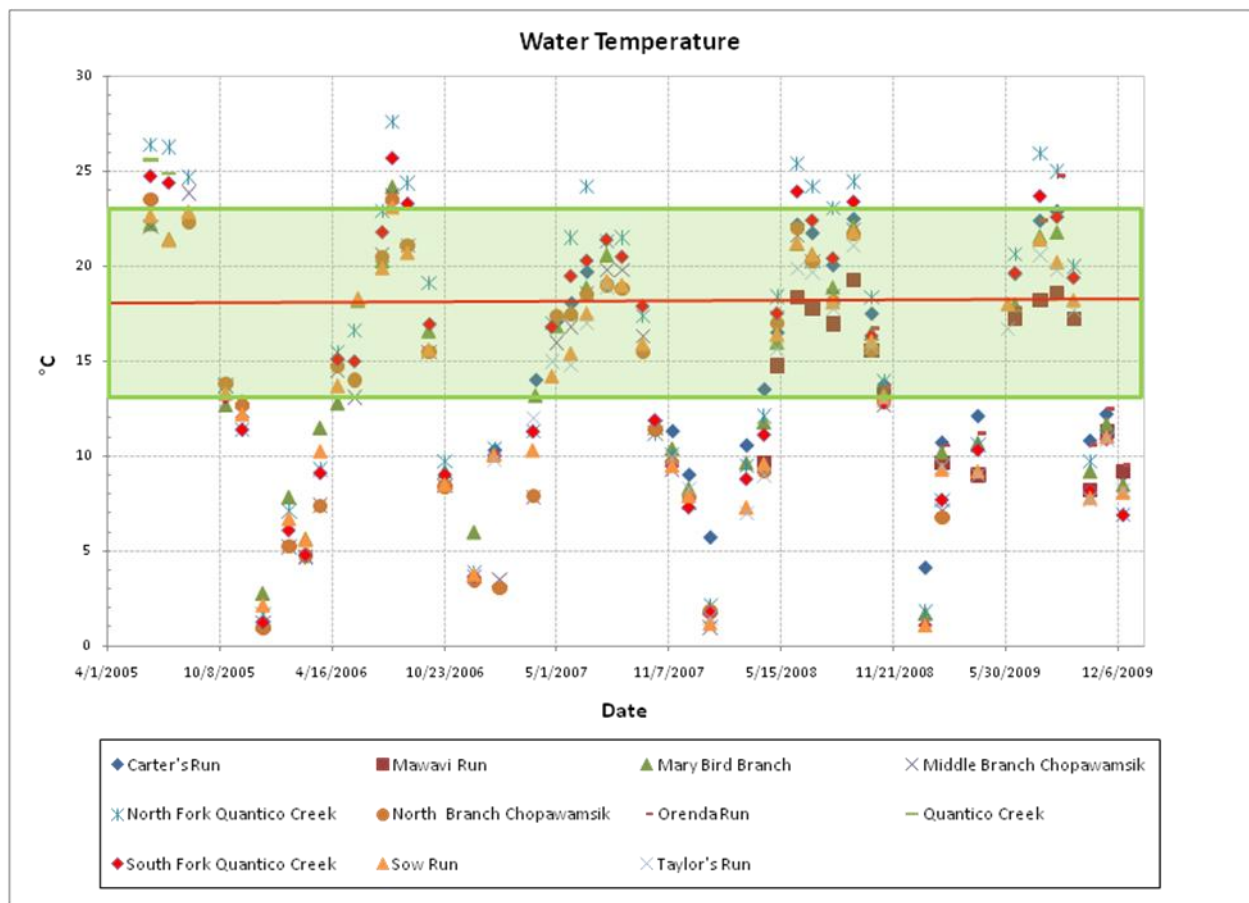
According to Old (1932) “the general opinion of workers (although none mentioned it directly) as gleaned from the literature, is that sponges are very sensitive to alterations in their environment. The experience of the writer in attempts to culture sponges in the laboratory also seemed to point to a high selectivity.” Presumably water quality parameters affect the ability of *Ephydatia muelleri* to survive in a stream, because the sponge is a sessile organism and is unable to migrate to areas with more favorable conditions. Old (1932) goes on to describe the ideal habitat for *E. Muelleri* as “smooth streams, open to sunlight; bottom soil, sand, gravel, with or without organic deposits; slope of adjacent land, pronounced to gradual; surroundings, natural. Water: pollution absent; turbidity 7 or less; color 20-80; earthy, grassy, musty odor; summer temperatures 61-75 F, pH 7.1 – 8.0.” According to Lauer et al. (2001) freshwater sponge distribution is limited by calcium, silicon, bound carbon dioxide and pH. Growth is dictated by temperature (see Figure 5).

**Table 3: Range of physical and chemical characteristics of perennial streams in Quantico Creek watershed in Prince William Forest Park (NCRN) 2005-2010, compared to published ideal habitat for *E. muelleri*.**

		Ideal Conditions				PRWI Streams							
		Old 1932	Jewell 1935	Lauer et al. 2001	Smith 2001	Mawavi Run	Sow Run	Taylor Run	Carters Run	Mary Bird Branch	Orenda Run	South Fork Quantico Creek	North Fork Quantico Creek
<b>Nitrate</b>	mg/l			0.1-2.4		0.3-0.9	0.1-1.2	0.1-1.2	0.12-1.1	0.1-1.2	0.6-1.3	0.1-1.2	0-1.3
<b>Phosphorus</b>	mg/l			0.01-0.24		0.1-0.47	0.11-4.95	0.11-1.85	0.09-1.63	0.08-5.87	0.12-0.37	0.04-5.21	0.08-5.89
<b>pH</b>		7.1-8.0	6.6-8.4	7.7-8.4	>7.0	6.0-7.3	6.5-7.5	6.8-7.4	5.9-6.6	6.5-10.4	6.3-7.0	6.4-13.0	6.4-7.3
<b>Specific conductance</b>	µS/cm		38-94	288-577		34-66	21-474	0-115	0-106	0-67	100-152	51-87	56-686
<b>Water Depth</b>	ft	0.1-1.0				0.3-0.6	0.0- 1.0	0.0-1.0	0.0-0.3	0.0-0.5	0.0-0.2	0.2-1.5	0. -1

PRWI streams in Tables 3 are listed in order from upstream (left) to downstream (right). Streams on the south side of South Fork Quantico Creek (Carters Run, Mawavi Run, and Orenda Run) have lower pH and ANC than those on the north (Mary Bird Branch, North Fork Quantico Creek, Sow Run, and Taylor Run). This difference is presumably due to a difference in geology and soils.

*Ephydatia muelleri* prefers alkaline environments (above 7.0 pH) (Smith 2001). Jewell (1935) found *Ephydatia muelleri* in Wisconsin streams ranging from 6.6 to 7.6 pH (green rectangle), 7.1 – 8.0 (Old 1932). Based on observed pH values (Table 3), we would not expect to find *Ephydatia muelleri* in Carter’s Run or Mawavi Run. Sponges have not been observed in these streams.



**Figure 5: Stream Temperature in Prince William Forest Park**

Cell division and migration is referred to as germination. Germination and subsequent hatching of gemmules occur based on conditions such as temperature (Simpson and Fell 1974). Upon gemmule hatching, new sponges are formed. Germination of freshwater sponges occurs between 13 °C and 23 °C requiring two weeks of sustained temperatures in autumn and a few days in spring (Smith 2001). Temperatures between 13 °C and 23 °C (green shading) in PRWI occur May through September, sometimes including April or October.

In the Danube floodplain, in Europe, *Ephydatia muelleri* significantly avoided water bodies with temperatures less than 17.5 °C (Droescher and Waringer 2007). In PRWI, water temperatures above 17.5 (red line) occur late May through October, ideal sponge growing season. Old (1932) describes 16 °C to 24 °C as ideal *E. muelleri* habitat.

## Discussion

### Classification

Kingdom: *Animalia*

Phylum: *Porifera*

Class: *Demospongia*

Order: *Haplosclerida*

Family: *Spongillidae*

Genus: *Ephydatia*

Species: *muelleri*

The majority of sponges (Porifera) are found in marine environments. Only 1 family (Spongillidae) occurs in freshwaters of the United States (Smith 2001). The genus *Ephydatia* was described in 1816 by Lamouroux, and the species *muelleri*, by Lieberkuhn in 1855 (Penny and Racek 1968). Its common name is “Mueller’s freshwater sponge.” *Ephydatia muelleri* is distributed throughout the Northern Hemisphere in temperate regions including Europe, North America and Asia (Thorp and Covich 2010).

Little is known about freshwater sponges in the National Capital Region and surrounding areas. Wisconsin DNR biologists “were unable to assess adequately the conservation status of sponges” during their cataloging of North American state and regional freshwater sponge references. A lack of research, specimens and literature are cited as the cause (Watermolen 2008). *E. muelleri* is found in temperate areas of North America, however information available on the occurrence of sponges does not match their overall expected distribution (Thorp and Covich 2010). *E. muelleri* are reported in Central Europe, Connecticut (Paduano and Fell 1997), Michigan (Lauer et al. 2001), Wisconsin, Arizona (Sowka 1999), and Virginia (Cheryl Bright email to Klaus Ruetzler, 29 Oct 2009). *E. muelleri* is one of the four most common freshwater sponge species in the United States (Smith 2001).

The occurrences of unidentified species in Wolf Trap Creek at Wolf Trap National Park for the Performing Arts in 1975 and at Mine Run Branch near George Washington Memorial Parkway in 1975 & 1976 (1979 data from George Mason University, see Table 1) prompted a brief search by the water monitoring crew. During the search of Wolf Trap Creek and Mine Run Branch near NCRN water monitoring sites *Porifera* were not observed. In October of 1974, there was also an observation in Pimmit Run between George Washington Memorial Parkway and the Potomac River. Pimmit Run has not been surveyed for sponges. We believe their presence is unlikely due to the presence of heptachlor epoxide and chlordane above human contact thresholds and because freshwater sponges are found in water bodies with high quality water and low levels of pollutants (Holley 2009).

### Ecology

Freshwater sponges are sessile, benthic filter feeding invertebrates that live in freshwater streams and ponds and reproduce both sexually and asexually. Their geographic range includes North America, Europe and Asia (Thorp and Covich 2010). Sponges will grow on upper, side or lower surfaces of any stable submerged substrate (rocks, pebbles, aquatic vegetation, logs, branches and twigs) in clean ponds, lakes, streams and rivers. In South Fork Quantico Creek, sponges

have been observed growing on a rusted metal grate submerged in the stream. In very favorable habitat sponge growth can obscure surfaces and clog pipes, but this is rare (Smith 2001). Habitat conditions, such as water current, available light and depth, affect the appearance of freshwater sponges. Because of these factors, the morphology can vary within the same species. The physical characteristics of their growth patterns include branched, clumped and encrusted forms (Thorp and Covich 2010). *Porifera* can range in growth form from a simple encrustation to tufts of long fingerlike projections and in color from white to yellow or green (Smith 2001).

Reproduction in freshwater sponges can occur both sexually and asexually. One method of asexual reproduction is fragmentation in which pieces break off and are able to regenerate and grow independently. Another method of asexual reproduction is gemmule formation. Gemmules are reproductive, ball-shaped masses of cells (Figure 4) surrounded by a protective coating. Gemmule formation is specific to freshwater sponges and does not occur in marine sponges (Potts et al. 2009). Most temperate freshwater sponge species go dormant during seasonal fluctuation of temperatures, particularly low winter temperatures that freeze streams (diapause). They also go dormant due to extremely high temperatures and environmental stress (Thorp and Covich 2010).

Gemmules are produced before the sponges enter diapause and are able to survive overwinter (Hill and Hill 2002). Gemmules may remain attached to the substrate or become freed and rise to the surface or sink to the bottom. They overwinter easily, can withstand repeated freezing and thawing, and can be viable after three years of drying (Smith 2001). Once temperatures warm (over 4°C, although gemmules will slowly hatch even at 3°C) and are maintained, hatching will begin. The gemmule cells differentiate and form an active sponge (Thorp and Covich 2010). Germination occurs in water of 13°C to 23°C, requiring two weeks or more in autumn and a few days in the spring (Smith 2001). Gemmules of *Ephydatia muelleri* are able to withstand environmental anoxia for at least four months independent of temperature within the normally encountered range of this species (Reiswig and Miller 1998). It is possible that gemmules of this and other freshwater sponges buried in the top layers of anoxic sediments constitute “gemmule banks” that may have the potential to survive for many years. While gemmules of *Ephydatia muelleri* can survive dehydration for months, this subject has not been experimentally investigated for this species, although information is available for other species (De Santo and Fell 1996). Some freshwater sponge gemmules can remain dormant for longer than one winter (Caceres 1997).

### **Community Associations**

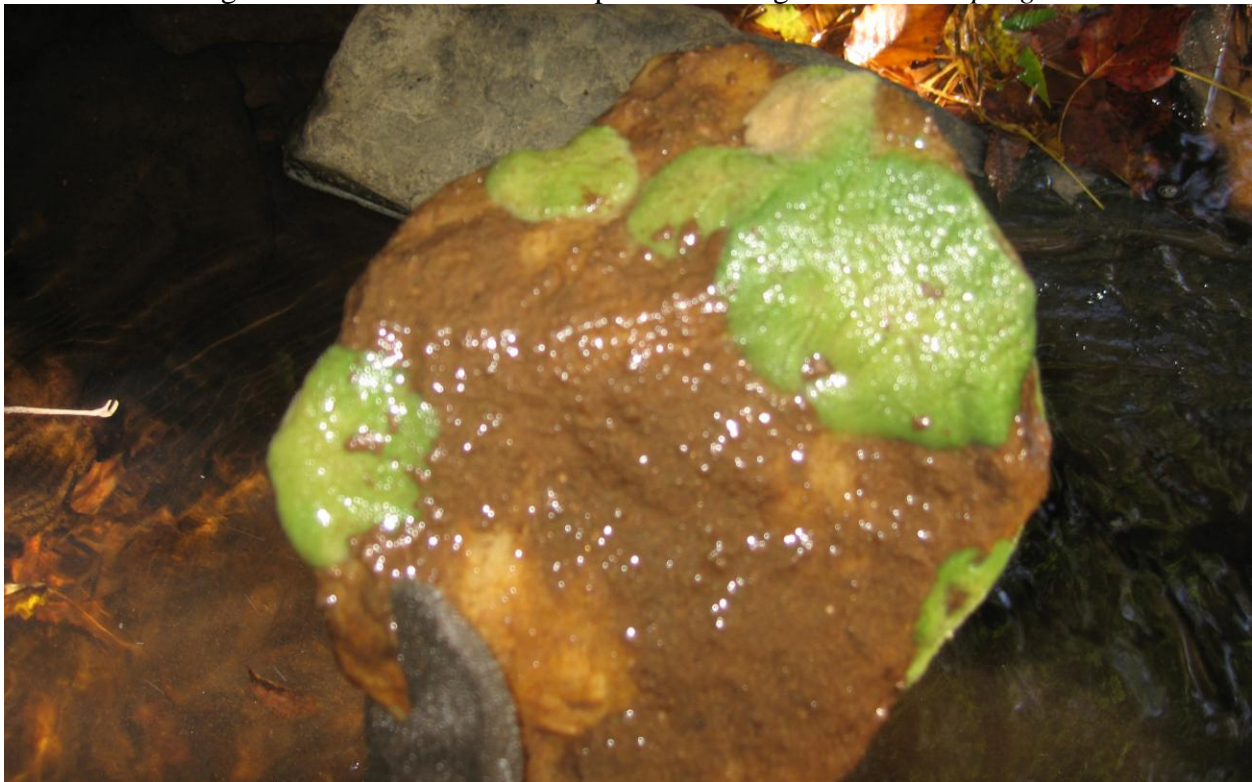
One may find up to three species of freshwater sponges in the same body of water, but rarely more. *E. muelleri* was found in the floodplain waters of Austria’s Danube River in association with *Eunapius fragilis* and *Spongilla lacustris*, where it preferred wood over stone substrate (Droescher and Waringer 2007). Lauer et al. (2001) reported the three species to be growing in close proximity in some cases touching or overlapping. They also reported that the three species have similar preferences for water quality characteristics and are often found growing together (Jewell 1935, Smith 2001). This association is commonly reported in other parts of North America. Jewell (1935) noted strong associations of *Ephydatia muelleri* with *Spongilla fragillilis* and *Spongilla lacustris*. She described “two distinct and mutually exclusive association groups”; consisting of (1) *Ephydatia muelleri* and *Spongilla fragillilis* and (2) *Ephydatia everetti* and *Spongilla lacustris*. She thought this indicated environmental factors necessary for one pair



prohibited the other pair. *Eunapius fragilis* and *Spongilla lacustris* are at the opposite end of the silicon and hydrological connectivity spectrum and therefore do not seem to be competitive with *E. muelleri* (Lauer et al. 2001).

“Areal cover of sponges may better indicate the benthic community organization, as resource limitations are often spatially distributed among sessile benthic invertebrates (Connell 1961, Jackson and Buss 1975, Jackson 1979, Russ 1982, Ricciardi et al. 1995, Lauer and Spacie 2000). This would imply areal cover by sponges is more important than colony number or morphometry. Colony size changes dramatically on an annual basis. Sponges typically begin vegetative growth in the spring or early summer and increase in size throughout the autumn. A reduction of body tissue and the formation of gemmules in the fall allow the sponge to overwinter with the cycle starting over the following spring (Frost et al 1982, Pennak 1989)” (Lauer et al. 2001).

Freshwater sponges can be an important component of stream ecosystem processes (Thorp and Covich 2010). Freshwater sponges feed via a combination of filtration of organic particles and photosynthesis of endosymbiotic *Zoochlorellae* (Sand-Jensen and Pendersen 1994). Green sponges (Figure 6) indicate presence of *Zoochlorellae*, a facultative symbiotic algae. Sponges that appear green get their color from the presence of symbiotic algae (Sand-Jensen and Pendersen 1994). The algal-invertebrate relationship results in mixotrophic nutrition in which autotrophic and heterotrophic processes are combined. *Spongillidae* benefits the alga by providing nutrients such as nitrogen, phosphorous and carbon dioxide. The sponge benefits by consuming the carbohydrates produced by the photosynthesizing algae (Thorp and Covich 2010). Though it has not yet been positively identified, the likelihood of the algae being *Zoochlorellae* is good because of its common presence in green colored *Spongillidae*.



**Figure 6: *Ephydatia muelleri* encrusted on a rock taken out of the stream. It appears green due to the symbiotic algae *Zoochlorellae*.**

Larvae of the caddis fly, *Ceraclea fluva*, build their case by feeding on the sponge and using its siliceous spicules (Corallini and Gaino 2003). Freshwater sponges are also food for ducks, crayfish, aquatic insects (DeSanto Elizabeth and Fell 1996) and zooplankton (laboratory observation, 27 Oct 2009) and feed on bacterio-plankton (Ruetzler, K. email to Tonya Watts. 28 Oct.2009).

## Recommendations

It is recommended that the extent of freshwater sponges in Prince William Forest Park be determined. Additionally, the symbiotic algae in the known colonies of *Ephydatia muelleri* should be identified in order to confirm if it is *Zoochlorellae*.

Additional study of freshwater sponges in the National Capital Region is highly recommended. These are to include an inventory of the sponges to determine if other species occur, as well as, chemical and physical characterization of stream locations where the sponges are found to determine the conditions allowing their presence.

Because little is known about the presence and distribution of freshwater sponges, not only in the Washington DC metropolitan area but across the United States, this report should be widely distributed among natural resource professionals within the National Park Service, as well as our water monitoring colleagues in the surrounding area.

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